IN THE UNITED STATES PATENT AND TRADEMARK OFFICE

In re application of:	
HARTLEY *et al.*	Art Unit: (To be assigned)
Appl. No.: (To be assigned; Rule 53(b) Continuation of 09/432,085)	Examiner: (To be assigned)
Filed: (Herewith)	Atty Docket: 0942.285000H/RWE/BJD
For: **Recombinational Cloning Using Engineered Recombination Sites**	

Preliminary Amendment and Remarks

Commissioner for Patents
Washington, DC 20231

Sir:

In advance of prosecution in the above-identified application, Applicants submit the following amendments and remarks. This Preliminary Amendment and Remarks is provided in the following format:

(A) A clean version of each replacement paragraph/section/claim along with clear instructions for entry;

(B) Starting on a separate page, appropriate remarks. *See* 37 C.F.R. § 1.121 and MPEP § 714; and

(C) Starting on a separate page, a marked-up version entitled: "Version with markings to show changes made."

It is not believed that extensions of time or fees for net addition of claims are required beyond those that may otherwise be provided for in documents accompanying this paper. However, if additional extensions of time are necessary to prevent abandonment of this application, then such extensions of time are hereby petitioned under 37 C.F.R. § 1.136(a), and

any fees required therefor (including fees for net addition of claims) are hereby authorized to be charged to our Deposit Account No. 19-0036.

Amendments

Please amend the application as follows:

In the Specification:

In the specification at page 1, please delete the paragraph appearing at lines 4-7 (the cross-reference paragraph) and insert therefor the following paragraph:

CROSS REFERENCE TO RELATED APPLICATIONS

The present application is a continuation of U.S. Application No. 09/432,085, filed November 2, 1999, which is a divisional of U.S. Application No. 09/233,493, filed January 20, 1999 (now U.S. Patent No. 6,143,557), which is a continuation of U.S. Application No. 08/663,002, filed June 7, 1996 (now U.S. Patent No. 5,888,732), which is a continuation-in-part of U.S. Application No.08/486,139, filed June 7, 1995 (now abandoned), which applications are entirely incorporated herein by reference.

In the specification at page 2, please delete the paragraph appearing at lines 12-14 in its entirety.

In the specification at page 51, please delete the paragraph appearing at lines 1-12 and substitute therefor the following paragraph:

Part V: Expression of Fusion Proteins

Two colonies from each transformation were picked into 2 ml of rich medium (CIRCLEGROW® brand culture medium, Bio101 Inc.) in 17 × 100 mm plastic tubes (FALCON® brand plasticware, Cat. No. 2059, Becton Dickinson) containing 100 μg/ml ampicillin and shaken vigorously for about 4 hours at 37°C, at which time the cultures were visibly turbid. One ml of each culture was transferred to a new tube containing 10 μl of 10% (w/v) IPTG to induce expression of GST. After 2 hours additional incubation, all cultures had about the same turbidity; the A600 of one culture was 1.5. Cells from 0.35 ml each culture were harvested and treated with sample buffer (containing SDS and β-mercaptoethanol) and aliquots equivalent to about 0.15 A600 units of cells were applied to a Novex 4-20% gradient polyacrylamide gel. Following electrophoresis the gel was stained with Coomassie blue.

In the Claims:

Please cancel claims 2-34, without prejudice to or disclaimer of the subject matter contained therein.

Remarks

The foregoing amendments to the specification have been made to update the priority information at page 1 of the specification, to delete text at page 2 of the specification, and to provide proper format for the trademarks used in the application at pages 70 and 77 in accordance with MPEP § 608.01(v). Accordingly, the foregoing amendments do not add new matter, and their entry into the present application is respectfully requested.

In accordance with 37 C.F.R. § 1.821, the paper and computer-readable copies of the sequence listing included herewith are the same.

Applicants believe that the present application is now in condition for examination. If the Examiner believes, for any reason, that personal communication will expedite prosecution of this application, the Examiner is invited to telephone the undersigned at the number provided.

Prompt and favorable consideration of the foregoing amendments, and entry of the same into the present application, are respectfully requested.

Respectfully submitted,

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P108-17.wpd

Version with markings to show changes made

In the Specification:

In the specification at page 1, the paragraph appearing at lines 4-17 (the Cross-Reference section) is sought to be amended as follows:

[***Cross-Reference to Related Applications***]

CROSS-REFERENCE TO RELATED APPLICATIONS

The present application is a continuation of U.S. Application No. 09/432,085, filed November 2, 1999, which is a divisional of U.S. Application No. 09/233,493, filed January 20, 1999 (now U.S. Patent No. 6,143,557), which is a continuation of U.S. Application No. 08/663,002, filed June 7, 1996 (now U.S. Patent No. 5,888,732), which is a continuation-in-part of U.S. [Appl.] Application No.08/486,139, filed June 7, 1995 (now abandoned), which [application is] applications are entirely incorporated herein by reference.

In the specification at page 2, the paragraph appearing at lines 12-14 is sought to be deleted.

In the specification at page at page 51, the paragraph appearing at lines 1-12 is sought to be amended as follows:

Part V: Expression of Fusion Proteins

Two colonies from each transformation were picked into 2 ml of rich medium [(CircleGrow] (CIRCLEGROW® brand culture medium, Bio101 Inc.) in 17 × 100 mm plastic tubes [Falcon 2059] (FALCON® brand plasticware, Cat. No. 2059, Becton Dickinson) containing 100 μg/ml ampicillin and shaken vigorously for about 4 hours at 37°C, at which time the cultures were visibly turbid. One ml of each culture was transferred to a new tube containing 10 μl of 10% (w/v) IPTG to induce expression of GST. After 2 hours additional incubation, all cultures had about the same turbidity; the A600 of one culture was 1.5. Cells from 0.35 ml each culture were harvested and treated with sample buffer (containing SDS and β-mercaptoethanol) and aliquots equivalent to about 0.15 A600 units of cells were applied to a Novex 4-20% gradient polyacrylamide gel. Following electrophoresis the gel was stained with Coomassie blue.